Isomers in Commercial Samples of Conjugated Linoleic Acid

Sir:

In connection with our analytical activities (MRS Lipid Analysis Unit), we have analyzed several commercial samples of conjugated linoleic acid (CLA). Most of these have been prepared by alkali-isomerization of linoleic acid or of oils, such as sunflower or safflower, rich in this acid. (i) We have examined the methyl esters, which must be prepared from the acids avoiding the use of acidic catalysts, by gas chromatography (GC). With a 25-m carbowax capillary column, most of the samples show two major peaks which are well-resolved from each other, along with minor peaks running later which are first the all-*cis* and then the all-*trans* dienes. The two major peaks are usually considered to be only the 9*c*,11*t* and 10*t*,12*c* isomers. This interpretation is not consistent with the GC/mass spectrometry (MS) data reported below. When examined on a highly polar 100-m capillary column (CP Sil 88), the GC trace is more complex. Several samples show a new peak for 11,13 diene, and some indicate the presence of several other isomers. We also have evidence that the 9,11 peak contains the 8,10 isomer though we have been unable to resolve these. (ii) GC–MS of the diene adducts formed through reaction with 4-methyl-2,3,4-triazoline-3,5-dione (MTAD derivatives), with selected ion monitoring, shows the presence of 8,10; 9,11; 10,12; and 11,13 dienes (1). For example, Figure 1 illustrates the selected ion chromatogram for one of the diagnostic ions from each isomer in a commercial CLA preparation. It is clear that four unresolved isomers are present. The proportions of these vary widely (presumably depending on the conditions of alkali-isomerization) and even more isomers are sometimes present. GC–MS of the dimethyloxazoline derivatives confirmed the identity of the major products. (iii) High-resolution ^{13}C nuclear magnetic resonance spectroscopy confirmed the presence of at least four *cis,trans* conjugated dienes and gave quantitative results in line with those obtained by GC–MS analysis.

We conclude that most samples of CLA, though rich in the 9,11 (probably mainly if not entirely the 9*c*,11*t* isomer) and 10,12 dienes (probably mainly if not entirely the 10*t*,12*c* isomer) also contain at least the 8,10 and 11,13 *cis,trans* dienes, sometimes at quite high levels. These are accompanied by all-*cis* and all-*trans* dienes. In one commercial sample of CLA, we found the following dienes: 8,10 (14%), 9,11 (30%), 10,12 (31%), and 11,13 (24%).

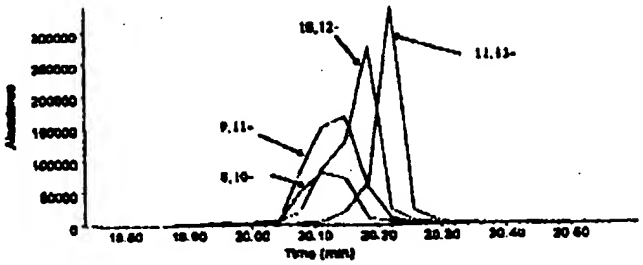

FIG. 1. Selected ion scan in gas chromatography–mass spectrometry of methyl triazoline dione adducts of a commercial conjugated linoleic acid preparation (methyl esters). The characteristic ions are at *m/z* = 264 (8,10 18:2), 250 (9,11–18:2), 236 (10,12–18:2), and 222 (11,13–18:2).

It is important that those who produce these materials and use them for research purposes appreciate the complex nature of their products. To our knowledge, the identity of the biologically active CLA is not known although it is generally assumed to be 9*c*,11*t*-18:2. Nor is it known how the activity of this isomer may be influenced by the presence of other isomeric CLA.

REFERENCE

1. Dobson, G., Identification of Conjugated Fatty Acids by Gas Chromatography–Mass Spectrometry of 4-Methyl-1,2,4-triazoline-3,5-dione adducts, *J. Am. Oil Chem. Soc. 75*, Jan. 1998, in press.

William W. Christie*,
Gary Dobson, and
Frank D. Gunstone,
Scottish Crop Research Institute,
Invergowrie,
Dundee DD5 2DA,
Scotland

[Received August 29, 1997; accepted September 15, 1997]

*To whom correspondence should be addressed.
E-mail: wchris@scri.sari.ac.uk.